

Nika Adham, et al.
Serial No.: 09/116,676
Filed: July 16, 1998
Page 2

E1
Cont
embodiment, the DNA is cDNA. In another embodiment, the DNA is genomic DNA. In another embodiment, the nucleic acid is RNA. In another embodiment, the nucleic acid is linked to a nucleic acid encoding a FLAG® epitope. --

Please delete the paragraph on page 62, lines 13-26, and insert the following replacement paragraph:

E2
--Recombinant baculovirus expressing hOb-Re was generated using the Bac-to-Bac Baculovirus Expression System (Gibco BRL). A SacI/HindIII fragment containing the entire encoding region of hOb-Re was isolated from B025, and ligated to the vector pFASTBacI at the SacI and HindIII sites. A second construct was made, using a Chameleon Double-Stranded Site-Directed Mutagenesis Kit (Stratagene), which differed in that it contained an 8 amino acid FLAG® epitope (Kodak) downstream of the signal sequence cleavage site in hOb-Re. Recombinant bacmid E.coli colonies were generated and DNA for each was isolated as described by the manufacturer. The constructs with and without the FLAG® epitope are designated B047 and B045, respectively. --

In the Claims:

Please cancel claims 1-9, 14-48, 150, 208 and 213 without disclaimer or prejudice to applicants' right to pursue the subject matter of these claims in a continuation or divisional application.

Please amend claims 222, 224, 226 and 228 as follows:

E3 Sub 31
--222. (Three Times Amended) A method of obtaining a composition which comprises:

(a) identifying a chemical compound by the process of

E3
Cont
Sub
31
cont

any of claims 224, 209, 225, 226, 214 or 227, or a structural and functional analog or homolog thereof,

- (b) recovering said chemical compound free of any soluble polypeptide, and
- (c) admixing a carrier and a pharmaceutically effective amount of said chemical compound.

-224. (Amended) A process for determining whether a chemical compound specifically binds to:

- (a) a soluble polypeptide comprising consecutive amino acids, the amino acid sequence of which is shown in Figure 5 (SEQ ID NO: 10);
- (b) a soluble polypeptide having a sequence which varies therefrom by no more than 15 amino acids, such variations:
 - (i) not involving amino acids corresponding to the amino acids at positions 799-804 of the amino acid sequence shown in Figure 5 (SEQ ID NO: 10); and
 - (ii) not changing the functional properties of the soluble polypeptide; or
- (c) a soluble polypeptide comprising the soluble polypeptide of (a) or (b) linked to consecutive amino acids corresponding to a FLAG® epitope,

E4

which comprises contacting the soluble polypeptide of (a), (b) or (c) above with the compound under conditions suitable for binding, and detecting

E⁴
Cont.

specific binding of the chemical compound to the
soluble polypeptide. --

-226. (Amended) A process involving competitive binding for
determining whether a first chemical compound
specifically binds to:

- (a) a soluble polypeptide comprising consecutive
amino acids, the amino acid sequence of
which is shown in Figure 5 (SEQ ID NO: 10);
- (b) a soluble polypeptide having a sequence
which varies therefrom by no more than 15
amino acids, such variations:
 - (i) not involving amino acids corresponding
to the amino acids at positions 799-804
of the amino acid sequence shown in
Figure 5 (SEQ ID NO: 10); and
 - (ii) not changing the functional properties
of the soluble polypeptide; or
- (c) a soluble polypeptide comprising the soluble
polypeptide of (a) or (b) linked to
consecutive amino acids corresponding to a
FLAG[®] epitope,

which comprises contacting the soluble polypeptide of
(a), (b) or (c) above, with both the first chemical
compound and a second chemical compound known to bind
to the soluble polypeptide, and separately with only
the second chemical compound, under conditions
suitable for binding of both the first and second
compounds, and detecting specific binding of the first

E⁵

E5
Cont

chemical compound to the soluble polypeptide, a decrease in the binding of the second chemical compound to the soluble polypeptide in the presence of the first chemical compound indicating that the first chemical compound binds to the soluble polypeptide. --

-228. (Amended) A method of screening a plurality of chemical compounds not known to bind to:

- E6
- (a) a soluble polypeptide comprising consecutive amino acids, the amino acid sequence of which is shown in Figure 5 (SEQ ID NO: 10);
 - (b) a soluble polypeptide having a sequence which varies therefrom by no more than 15 amino acids, such variations:
 - (i) not involving amino acids corresponding to the amino acids at positions 799-804 of the amino acid sequence shown in Figure 5 (SEQ ID NO: 10); and
 - (ii) not changing the functional properties of the soluble polypeptide; or
 - (c) a soluble polypeptide comprising the soluble polypeptide of (a) or (b) linked to consecutive amino acids corresponding to a FLAG[®] epitope,

to determine whether a compound specifically binds to the soluble polypeptide of (a), (b) or (c) which comprises:

- (1) preparing a cell extract or cell supernatant from

cells transfected with and expressing DNA encoding the soluble polypeptide and contacting the cell extract or cell supernatant with a compound known to bind specifically to the soluble polypeptide;

- El
Cont
- (2) contacting the preparation of step (1) with the plurality of compounds not known to bind specifically to the soluble polypeptide, under conditions permitting binding of compounds known to bind the soluble polypeptide;
 - (3) determining whether the binding of the compound known to bind to the soluble polypeptide is reduced in the presence of the compounds, relative to the binding of the compound in the absence of the plurality of compounds; and if so
 - (4) separately determining the binding to the soluble polypeptide of each compound included in the plurality of compounds, so as to thereby determine whether a compound specifically binds to the soluble polypeptide of (a), (b) or (c).--
-

Applicants submit hereto as **Exhibit A** a marked up version of the amendments showing the changes made.

REMARKS

Claims 1 to 9, 14 to 48, 150, 208, 209, 213, 214, 218-220 and 222-228 were pending in the subject application. By this Amendment, applicants have canceled claims 1-9, 14-48, 150, 208, and 213 without prejudice or disclaimer, and have amended claims